6. (Reiterated) The method according to Claim 23, wherein the gene encoding said endogenous extracellular protease has been deleted by homologous or illegitimate recombination.

7. (Reiterated) The method according to Claim 23, wherein a plasmid comprises said expression cassette.

9. (Reiterated) The method according to Claim 7, wherein said mutant high alkaline protease is obtained from *Bacillus* novo species PB92.

10. (Reiterated) The method according to Claim 23, wherein at least one copy of said expression cassette is integrated into the genome of said host.

11. (Reiterated) The method according to Claim 10, wherein said host further contains at least one copy of a plasmid comprising said expression cassette.

12. (Reiterated) A method of obtaining an alkalophilic *Bacillus* strain having no detectable extracellular high alkaline protease, said method comprising:

transforming an alkalophilic *Bacillus* strain with a cloning vector comprising the 5' and the 3' flanking regions but not the coding region of gene coding for the high alkaline protease and encoding a replication function, wherein a sufficient amount of said flanking regions is present to provide for homologous recombination with an endogenous gene coding for the high alkaline protease whereby transformants are obtained;

growing said transformants under growth conditions to which the replication function of said cloning vector is sensitive whereby the replication function encoded by said vector is inactivated; and

isolating said transformants identified as having said inactivated replication function and no detectable extracellular high alkaline protease.

13. (Amended) The method according to Claim 12, wherein said alkalophilic *Bacillus* strain is *Bacillus* novo species PB92 or a derivative thereof which is incapable of reversion and contains a mutant high alkaline protease.

14. (Amended) An alkalophilic Bacillus strain producing a mutant high alkaline protease which is [substantially] free of expression product of an indigenous extracellular alkaline protease gene, wherein said strain has been obtained by transforming an alkalophilic *Bacillus* strain having no detectable indigenous extracellular high alkaline protease obtained by the method according to Claim 12, 13, or 27 with a plasmid expression vector comprising the mutant high alkaline protease gene.

15. (Reiterated) The *Bacillus* strain according to Claim 14, wherein said alkalophilic

Bacillus strain is a mutant of *Bacillus* novo species PB92 or a derivative thereof.

19. (Twice Amended) A detergent composition comprising as an active ingredient at least one [or more] mutant [forms] form of high alkaline protease[, wherein at least one of a said mutant form of high alkaline protease has been] prepared according to the method of Claim 23.

23. (Twice Amended) A method for production of a mutated high alkaline protease [substantially] free of endogenous extracellular high alkaline protease, said method comprising:

growing an alkalophilic *Bacillus* strain host [substantially] incapable of reversion and having no detectable endogenous extracellular protease as a result of deletion of the gene for endogenous extracellular protease transformed with an expression cassette providing for expression of a said mutant high alkaline protease in said host, whereby said mutant high alkaline protease is produced; and

~~isolating said mutant high alkaline protease.~~

24. (Twice Amended) A method for preparing a detergent composition, which comprises the step of combining a detergent composition with, as an active ingredient, at least one [or more] mutant [forms] form of a high alkaline protease[, wherein at least one of a said mutant form of high alkaline protease has been] prepared according to the method of Claim 23.

25. (Twice Amended) A method for processing laundry, which comprises the step of contacting said laundry with a detergent composition comprising as an active ingredient at least one [or more] mutant [forms] form of a high alkaline protease[, wherein at least one of a said mutant form of high alkaline protease has been] prepared according to the method of Claim 23.

26. (Twice Amended) A method for production of a mutated high alkaline protease [substantially] free of endogenous extracellular protease, said method comprising:

growing an asporogenous *Bacillus* strain host having a reduced endogenous extracellular protease level as a result of deletion of the gene for said endogenous extracellular protease transformed with an expression cassette providing for expression of a mutated high alkaline protease in said host, whereby said mutated high alkaline protease is produced; and

isolating said mutant high alkaline protease.

27. (Reiterated) A method of obtaining an alkalophilic *Bacillus* strain having no detectable extracellular high alkaline protease, said method comprising:

transforming an alkalophilic *Bacillus* strain with a cloning vector comprising the 5' and the 3' flanking regions but not the coding region of gene coding for the high alkaline protease and wherein a sufficient amount of said flanking regions is present to provide for illegitimate recombination with an endogenous gene coding for the high alkaline protease whereby transformants are obtained;

growing said transformants under growth conditions to which the replication function of said cloning vector is sensitive whereby the replication function encoded by said vector is inactivated; and

isolating said transformants identified as having said inactivated replication function and no detectable extracellular high alkaline protease.

28. (Amended) A method for producing an alkalophilic asporogenic *Bacillus* novo species PB92 of [minimal] reduced endogenous extracellular protease level, said method comprising:

transforming an alkalophilic asporogenic *Bacillus* [strain] novo species PB92 with a specifically-mutated *Bacillus* novo PB92 alkaline protease.

29. (Amended) An alkalophilic *Bacillus* strain producing a mutant high alkaline protease which is [substantially] incapable of reversion and which is [substantially] free of expression product of an endogenous extracellular alkaline protease gene.

30. (Amended) A method for production of a mutated high alkaline protease [substantially] free of endogenous extracellular high alkaline protease, said method comprising:

isolating said mutant high alkaline protease from a culture broth or cell lysate of alkalophilic *Bacillus* strain host cells wherein said cells are substantially incapable of reversion and have no detectable endogenous extracellular protease as a result of deletion of the gene for endogenous extracellular protease and wherein said cells produce a mutant high alkaline protease as a result of transformation of said cells or predecessor cells with an expression cassette providing for expression of said mutant high alkaline protease in said host cells.

31. (Reiterated) The method according to Claim 30, wherein untransformed parent cells of said alkalophilic *Bacillus* strain host cells are *Bacillus novo* species PB92 strain cells or a derivative species thereof.

32. (Reiterated) The method according to Claim 30, wherein untransformed parent cells of said alkalophilic *Bacillus* strain host cells are asporogenic.

33. (Amended) The method according to Claim 30 wherein said alkalophilic *Bacillus*

strain host cells are [substantially] free of untransformed parent cells.

Add the following new claims:

--34. (New) An alkalophilic *Bacillus* strain comprising a non-reverting extracellular protease-negative phenotype, wherein said strain or an ancestor of said strain was stably transformed with an exogenous protease gene encoding a mutant high alkaline protease and wherein said strain has an increased efficiency in production of said mutant high alkaline protease as compared to an untransformed strain of the same species.

35. (New) The alkalophilic *Bacillus* strain according to Claim 34, wherein said phenotype is the result of a deletion of a sufficient amount of an endogenous extracellular protease so as to prevent reversion of said non-reverting extracellular protease-negative phenotype.

36. (New) A non-reverting mutant alkalophilic *Bacillus* strain comprising a mutated endogenous extracellular protease gene, wherein a sufficient amount of an endogenous extracellular protease gene has been deleted so as to prevent reversion of said strain when transformed with a mutated form of said exogenous extracellular protease gene.

37. (New) The non-reverting mutant alkalophilic *Bacillus* strain according to Claim 36, wherein reversion of said mutated form of said exogenous extracellular protease gene is prevented.--

REMARKS

The Present Invention

The present invention is directed to methods and compositions for preparation of mutant high alkaline proteases and non-reverting endogenous extracellular protease-negative alkalophilic and/or asporogenic *Bacillus* strains which produce the mutant high alkaline protease in the absence of the endogenous extracellular protease. The present invention is further directed to a detergent composition comprising as an active ingredient at least one mutant high alkaline proteases produced according to the method of the invention and a laundry process employing the detergent composition.